

REMARKS

Applicants filed the present Continuation application on June 28, 2001. In the Continuation application transmittal filed June 28, 2001, Applicants requested that claims 2-6, 8-13 and 40-66 be canceled before calculating the filing fee. Accordingly, claims 1, 7, and 14-39 are pending.

In the above amendment, claims 1, 7, and 14-29 have been canceled without prejudice. Claims 30-39 have been amended to recite the ATCC accession number for each respective hybridoma cell line. Claims 67-89 have been added. Support for added claims can be found on at least pages 8, 10, 11, 26-31, 40 and 41 of the specification.

The specification has also been amended. In particular, the pages of the specification providing the Brief Description of the Drawings has been amended to reflect the corresponding SEQ ID Nos in the Sequence Listing. The specification has also been amended to reflect the accession numbers and deposit dates assigned by the ATCC depository for Applicants' deposited biological materials. For the Examiner's convenience, a copy of the ATCC confirmation sheet providing the deposit information is enclosed herewith. The specification has further been amended to correct certain inadvertent typographical errors in various reference citations and correct the numbering of certain Examples. It is submitted that no new matter has been introduced by these amendments.

The amendments to the specification and claims are illustrated on the attached pages entitled "Marked Up Version to Show Changes Made".

For the Examiner's convenience, a clean copy of all the now pending claims 30-39 and 67-89 is provided above.

Respectfully submitted,

Date: August 13, 2001

By: Diane L. Marschang

Diane L. Marschang
Reg. No. 35,600

1 DNA Way
So. San Francisco, CA 94080-4990
Phone: (650) 225-5416
Fax: (650) 952-9881

Marked Up Version to Show Changes Made

On pages 2-3, in the paragraph on lines 31-42 and lines 1-6, the text has been amended as follows:

--Induction of various cellular responses mediated by such TNF family cytokines is believed to be initiated by their binding to specific cell receptors. Two distinct TNF receptors of approximately 55-kDa (TNFR1) and 75-kDa (TNFR2) have been identified [[Hohman] Hohmann et al., J. Biol. Chem., 264:14927-14934 (1989); Brockhaus et al., Proc. Natl. Acad. Sci., 87:3127-3131 (1990); EP 417,563, published March 20, 1991] and human and mouse cDNAs corresponding to both receptor types have been isolated and characterized [Loetscher et al., Cell, 61:351 (1990); Schall et al., Cell, 61:361 (1990); Smith et al., Science, 248:1019-1023 (1990); Lewis et al., Proc. Natl. Acad. Sci., 88:2830-2834 (1991); Goodwin et al., Mol. Cell. Biol., 11:3020-3026 (1991)]]. Extensive polymorphisms have been associated with both TNF receptor genes [see, e.g., Takao et al., Immunogenetics, 37:199-203 (1993)]. Both TNFRs share the typical structure of cell surface receptors including extracellular, transmembrane and intracellular regions. The extracellular portions of both receptors are found naturally also as soluble TNF-binding proteins [Nophar, Y. et al., EMBO J., 9:3269 (1990); and Kohno, T. et al., Proc. Natl. Acad. Sci. U.S.A., 87:8331 (1990)]. More recently, the cloning of recombinant soluble TNF receptors was reported by Hale et al. [J. Cell. Biochem. Supplement 15F, 1991, p. 113 (P424)]. --

On page 3, in the paragraph on lines 7-20, the text has been amended as follows:

--The extracellular portion of type 1 and type 2 TNFRs (TNFR1 and TNFR2) contains a repetitive amino acid sequence pattern of four

cysteine-rich domains (CRDs) designated 1 through 4, starting from the NH₂-terminus. Each CRD is about 40 amino acids long and contains 4 to 6 cysteine residues at positions which are well conserved [Schall et al., supra; Loetscher et al., supra; Smith et al., supra; Nophar et al., supra; Kohno et al., supra]. In TNFR1, the approximate boundaries of the four CRDs are as follows: CRD1- amino acids 14 to about 53; CRD2- amino acids from about 54 to about 97; CRD3- amino acids from about 98 to about 138; CRD4- amino acids from about 139 to about 167. In TNFR2, CRD1 includes amino acids 17 to about 54; CRD2- amino acids from about 55 to about 97; CRD3- amino acids from about 98 to about 140; and CRD4- amino acids from about 141 to about 179 [Banner et al., Cell, 73:431-[435] 445 (1993)]. The potential role of the CRDs in ligand binding is also described by Banner et al., supra. --

On page 3, in the paragraph on lines 21-40, text has been amended as follows:

--A similar repetitive pattern of CRDs exists in several other cell-surface proteins, including the p75 nerve growth factor receptor (NGFR) [Johnson et al., Cell, 47:545 (1986); Radeke et al., Nature, 325:593 (1987)], the B cell antigen CD40 [Stamenkovic et al., EMBO J., 8:1403 (1989)], the T cell antigen OX40 [Mallett et al., EMBO J., 9:1063 (1990)] and the Fas antigen [Yonehara et al., supra and Itoh et al., Cell, 66:233-243 (1991)]. CRDs are also found in the soluble TNFR (sTNFR)-like T2 proteins of the Shope and myxoma poxviruses [Upton et al., Virology, 160:20-29 (1987); Smith et al., Biochem. Biophys. Res. Commun., 176:335 (1991); Upton et al., Virology, 184:370 (1991)]. Optimal alignment of these sequences indicates that the positions of the cysteine residues are well conserved. These receptors are sometimes collectively referred to as members of the TNF/NGF receptor superfamily. Recent studies on p75NGFR showed that the deletion of CRD1 [Welcher, A.A. et al., Proc. Natl. Acad. Sci. USA, 88:159-163

(1991)] or a 5-amino acid insertion in this domain [Yan, H. and Chao, M.V., J. Biol. Chem., 266:12099-12104 (1991)] had little or no effect on NGF binding [Yan, H. and Chao, M.V., supra]. p75 NGFR contains a proline-rich stretch of about 60 amino acids, between its CRD4 and transmembrane region, which is not involved in NGF binding [Peetre, C. et al., Eur. J. [Hematol.] Haematol., 41:414-419 (1988); Seckinger, P. et al., J. Biol. Chem., 264:11966-11973 (1989); Yan, H. and Chao, M.V., supra]. A similar proline-rich region is found in TNFR2 but not in TNFR1.---

On page 6, in the paragraph on lines 38-39, the text has been amended as follows:

--Figure 1 shows the derived amino acid sequence of a native sequence DcR3 (SEQ ID NO:1). --

On page 6, in the paragraph on lines 41-42, the text has been amended as follows:

--Figure 2 shows the nucleotide sequence of a native sequence DcR3 cDNA (SEQ ID NO:2). --

On page 7, in the paragraph on lines 6-7, the text has been amended as follows:

--Figure 5 shows an alignment of DcR3 (SEQ ID NO:1) and human TNFR2 (hTNFR2) (SEQ ID NO:17). Four cysteine rich domains (CRD) are shown as CRD1, CRD2, CRD3, and CRD4. --

On page 7, in the paragraph on lines 9-10, the text has been amended as follows:

--Figure 6 shows an alignment of DcR3 (SEQ ID NO:3) and human OPG (SEQ ID NO:18). Four cysteine rich domains are identified as CRD1, CRD2, CRD3, and CRD4. --

On page 12, in the paragraphs on lines 24-37, the text has been amended as follows:

--Another means of increasing the number of carbohydrate moieties on the DcR3 polypeptide is by chemical or enzymatic coupling of glycosides to the polypeptide. Such methods are described in the art, e.g., in WO 87/05330 published 11 September 1987, and in Aplin and Wriston, CRC Crit. Rev. Biochem., vol. 10, pp. 259-306 (1981).

Removal of carbohydrate moieties present on the DcR3 polypeptide may be accomplished chemically or enzymatically or by mutational substitution of codons encoding for amino acid residues that serve as targets for glycosylation. Chemical deglycosylation techniques are known in the art and described, for instance, by [Hakimuddin] Sojar, et al., Arch. Biochem. Biophys., 259:52 (1987) and by Edge et al., Anal. Biochem., 118:131 (1981). Enzymatic cleavage of carbohydrate moieties on polypeptides can be achieved by the use of a variety of endo- and exo-glycosidases as described by Thotakura et al., Meth. Enzymol., 138:350 (1987). --

On page 13, in the paragraph on lines 16-27, the text has been amended as follows:

--Various tag polypeptides and their respective antibodies are well known in the art. Examples include poly-histidine (poly-his) or poly-histidine-glycine (poly-his-gly) tags; the flu HA tag polypeptide and its antibody 12CA5 [Field et al., Mol. Cell. Biol., 8:2159-2165 (1988)]; the c-myc tag and the 8F9, 3C7, 6E10, G4, B7 and 9E10 antibodies thereto [Evan et al., Molecular and Cellular Biology, 5:3610-3616 (1985)]; and the Herpes Simplex virus glycoprotein D (gD) tag and its antibody [Paborsky et al., Protein Engineering, 3(6):547-553 (1990)]. Other tag polypeptides include the Flag-peptide [Hopp et al., BioTechnology, 6:1204-1210 (1988)]; the KT3 epitope peptide [Martin et al., Science, 255:192-194 (1992)]; an α -tubulin epitope peptide [Skinner et al., J. Biol.

Chem., 266:[15163-15166] 14163-14166 (1991)]; and the T7 gene 10 protein peptide tag [Lutz-Freyermuth et al., Proc. Natl. Acad. Sci. USA, 87:6393-6397 (1990)]. --

On pages 14-15, in the paragraph on lines 42 and 1-15, the text has been amended as follows:

--Variations in the native full-length sequence DcR3 or in various domains of the DcR3 described herein, can be made, for example, using any of the techniques and guidelines for conservative and non-conservative mutations set forth, for instance, in U.S. Pat. No. 5,364,934. Variations may be a substitution, deletion or insertion of one or more codons encoding the DcR3 that results in a change in the amino acid sequence of the DcR3 as compared with the native sequence DcR3. Optionally the variation is by substitution of at least one amino acid with any other amino acid in one or more of the domains of the DcR3 molecule. The variations can be made using methods known in the art such as oligonucleotide-mediated (site-directed) mutagenesis, alanine scanning, and PCR mutagenesis. Site-directed mutagenesis [Carter et al., Nucl. Acids Res., 13:4331 [(1986)] (1985); Zoller et al., Nucl. Acids Res., 10:6487 (1982)], cassette mutagenesis [Wells et al., Gene, 34:315 (1985)], restriction selection mutagenesis [Wells et al., Philos. Trans. R. Soc. London SerA, 317:415 (1986)] or other known techniques can be performed on the cloned DNA to produce the DcR3 variant DNA. --

On pages 16-17, in the paragraph on lines 38-42 and 1-7, the text has been amended as follows:

--Suitable host cells for the expression of glycosylated DcR3 are derived from multicellular organisms. Examples of invertebrate cells include insect cells such as Drosophila S2 and Spodoptera Sf9, as well as plant cells. Examples of useful mammalian host cell lines include Chinese hamster ovary (CHO) and COS cells. More specific examples include monkey kidney CV1 line transformed by SV40

(COS-7, ATCC CRL 1651); human embryonic kidney line (293 or 293 cells subcloned for growth in suspension culture, Graham et al., J. Gen Virol., 36:59 (1977))Graham et al., J. Gen Virol., 36:59 (1977)); Chinese hamster ovary cells/-DHFR (CHO, Urlaub and Chasin, Proc. Natl. Acad. Sci. USA, 77:4216 (1980))Urlaub and Chasin, Proc. Natl. Acad. Sci. USA, 77:4216 (1980)); mouse sertoli cells (TM4, Mather, Biol. Reprod., 23:243-251 (1980))Mather, Biol. Reprod., 23:243-251 (1980)); human lung cells (W138, ATCC CCL 75); human liver cells (Hep G2, HB 8065); and mouse mammary tumor (MMT 060562, ATCC CCL51). The selection of the appropriate host cell is deemed to be within the skill in the art. --

On page 18, in the paragraphs on lines 8-36, the text has been amended as follows:

--An example of suitable selectable markers for mammalian cells are those that enable the identification of cells competent to take up the DcR3 nucleic acid, such as DHFR or thymidine kinase. An appropriate host cell when wild-type DHFR is employed is the CHO cell line deficient in DHFR activity, prepared and propagated as described by Urlaub et al., Proc. Natl. Acad. Sci. USA, 77:4216 (1980). A suitable selection gene for use in yeast is the *trp1* gene present in the yeast plasmid YRp7 [Stinchcomb et al., Nature, 282:39 (1979); Kingsman et al., Gene, 7:141 (1979); [Tschemper] Tschumper et al., Gene, 10:157 (1980)]. The *trp1* gene provides a selection marker for a mutant strain of yeast lacking the ability to grow in tryptophan, for example, ATCC No. 44076 or PEP4-1 [Jones, Genetics, 85:[12] 23-33 (1977)].

Expression and cloning vectors usually contain a promoter operably linked to the DcR3 nucleic acid sequence to direct mRNA synthesis. Promoters recognized by a variety of potential host cells are well known. Promoters suitable for use with prokaryotic hosts include the β -lactamase and lactose promoter systems [Chang et al., Nature, 275:[615] 617-624 (1978); Goeddel et al., Nature, 281:544 (1979)], alkaline phosphatase, a tryptophan (*trp*) promoter system [Goeddel, Nucleic Acids

Res., 8:4057 (1980); EP 36,776], and hybrid promoters such as the tac promoter [deBoer et al., Proc. Natl. Acad. Sci. USA, 80:21-25 (1983)]. Promoters for use in bacterial systems also will contain a Shine-Dalgarno (S.D.) sequence operably linked to the DNA encoding DcR3.

Examples of suitable promoting sequences for use with yeast hosts include the promoters for 3-phosphoglycerate kinase [Hitzeman et al., J. Biol. Chem., 255:[2073] 12073 (1980)] or other glycolytic enzymes [Hess et al., [J.] Adv. Enzyme Reg., 7:149 (1968); Holland, Biochemistry, 17:4900 (1978)], such as enolase, glyceraldehyde-3-phosphate dehydrogenase, hexokinase, pyruvate decarboxylase, phosphofructokinase, glucose-6-phosphate isomerase, 3-phosphoglycerate mutase, pyruvate kinase, triosephosphate isomerase, phosphoglucose isomerase, and glucokinase. --

On pages 22-23, in the paragraph on lines 36-42 and 1-18, the text has been amended as follows:

--Alternatively, non-human homologues of DcR3 can be used to construct a DcR3 "knock out" animal which has a defective or altered gene encoding DcR3 as a result of homologous recombination between the endogenous gene encoding DcR3 and altered genomic DNA encoding DcR3 introduced into an embryonic cell of the animal. For example, cDNA encoding DcR3 can be used to clone genomic DNA encoding DcR3 in accordance with established techniques. A portion of the genomic DNA encoding DcR3 can be deleted or replaced with another gene, such as a gene encoding a selectable marker which can be used to monitor integration. Typically, several kilobases of unaltered flanking DNA (both at the 5' and 3' ends) are included in the vector [see e.g., Thomas and Capecchi, Cell, 51:503 (1987) for a description of homologous recombination vectors]. The vector is introduced into an embryonic stem cell line (e.g., by electroporation) and cells in which the introduced DNA has homologously recombined with the endogenous DNA are selected [see e.g., Li et al., Cell, 69:915 (1992)]. The selected cells are then injected into a blastocyst of an animal (e.g., a mouse or rat) to form aggregation chimeras [see

e.g., Bradley, in *Teratocarcinomas and Embryonic Stem Cells: A Practical Approach*, E. J. Robertson, ed. (IRL, Oxford, 1987), pp. 113-[152] 151]. A chimeric embryo can then be implanted into a suitable pseudopregnant female foster animal and the embryo brought to term to create a "knock out" animal. Progeny harboring the homologously recombined DNA in their germ cells can be identified by standard techniques and used to breed animals in which all cells of the animal contain the homologously recombined DNA. Knockout animals can be characterized for instance, for their ability to defend against certain pathological conditions and for their development of pathological conditions due to absence of the DcR3 polypeptide. --

On pages 27-28, in the paragraph on lines 34-42 and lines 1-6, the text has been amended as follows:

--As described in the Examples below, anti-DcR3 monoclonal antibodies have been prepared. Several of these antibodies, referred to as 4C4.1.4; 5C4.14.7; 11C5.2.8; 8D3.1.5; and 4B7.1.1 have been deposited with ATCC and have been assigned deposit accession numbers HB-12573, HB-12574, HB-12572, HB-12571, and HB-12575, respectively, In one embodiment, the monoclonal antibodies of the invention will have the same biological characteristics as one or more of the antibodies secreted by the hybridoma cell lines deposited under accession numbers HB-12573, HB-12574, HB-12572, HB-12571 or HB-12575. The term "biological characteristics" is used to refer to the *in vitro* and or *in vivo* activities or properties of the monoclonal antibodies, such as the ability to bind to DcR3 or to substantially block Fas ligand/DcR3 binding. Optionally, the monoclonal antibody will bind to the same epitope as at least one of the antibodies specifically referred to above. Such epitope binding can be determined by conducting various assays, like those described herein and in the examples.--

On page 28, in the paragraph on lines 19-39, the text has

been amended as follows:

--The DcR3 antibodies of the invention may further comprise humanized antibodies or human antibodies. Humanized forms of non-human (e.g., murine) antibodies are chimeric immunoglobulins, immunoglobulin chains or fragments thereof (such as Fv, Fab, Fab', F(ab')₂ or other antigen-binding subsequences of antibodies) which contain minimal sequence derived from non-human immunoglobulin. Humanized antibodies include human immunoglobulins (recipient antibody) in which residues from a complementary determining region (CDR) of the recipient are replaced by residues from a CDR of a non-human species (donor antibody) such as mouse, rat or rabbit having the desired specificity, affinity and capacity. In some instances, Fv framework residues of the human immunoglobulin are replaced by corresponding non-human residues. Humanized antibodies may also comprise residues which are found neither in the recipient antibody nor in the imported CDR or framework sequences. In general, the humanized antibody will comprise substantially all of at least one, and typically two, variable domains, in which all or substantially all of the CDR regions correspond to those of a non-human immunoglobulin and all or substantially all of the FR regions are those of a human immunoglobulin consensus sequence. The humanized antibody optimally also will comprise at least a portion of an immunoglobulin constant region (Fc), typically that of a human immunoglobulin [Jones et al., Nature, 321:522-525 (1986); Riechmann et al., Nature, 332:323-[329] 327 (1988); and Presta, Curr. Op. Struct. Biol., 2:593-596 (1992)]. --

On page 29, in the paragraph on lines 13-19, the text has been amended as follows:

--Human antibodies can also be produced using various techniques known in the art, including phage display libraries [Hoogenboom and Winter, J. Mol. Biol., 227:381 [(1991)] (1992); Marks et al., J. Mol. Biol., 222:581 (1991)]. The techniques of

Cole et al. and Boerner et al. are also available for the preparation of human monoclonal antibodies (Cole et al., Monoclonal Antibodies and Cancer Therapy, Alan R. Liss, p. 77 (1985) and Boerner et al., J. Immunol., 147(1):86-95 (1991)). --

On page 29, in the paragraph on lines 26-35, the text has been amended as follows:

--Methods for making bispecific antibodies are known in the art. Traditionally, the recombinant production of bispecific antibodies is based on the co-expression of two immunoglobulin heavy-chain/light-chain pairs, where the two heavy chains have different specificities [Milstein and Cuello, Nature, 305:537-[539] 540 (1983)]. Because of the random assortment of immunoglobulin heavy and light chains, these hybridomas (quadromas) produce a potential mixture of ten different antibody molecules, of which only one has the correct bispecific structure. The purification of the correct molecule is usually accomplished by affinity chromatography steps. Similar procedures are disclosed in WO 93/08829, published 13 May 1993, and in Traunecker et al., EMBO J., 10:3655-3659 (1991). --

On page 30, in the paragraphs on lines 6-36, the text has been amended as follows:

--Heteroconjugate antibodies are also within the scope of the present invention. Heteroconjugate antibodies are composed of two covalently joined antibodies. Such antibodies have, for example, been proposed to target immune system cells to unwanted cells [U.S. Patent No. 4,676,980], and for treatment of HIV infection [WO 91/00360; [WO 92/200373] WO 92/20373; EP 03089]. It is contemplated that the antibodies may be prepared *in vitro* using known methods in synthetic protein chemistry, including those involving crosslinking agents. For example, immunotoxins may be constructed using a disulfide exchange reaction or by forming a thioether bond. Examples of suitable reagents for this purpose include iminothiolate and methyl-4-mercaptobutyrimidate and those disclosed, for example, in U.S. Patent No. 4,676,980.

F. Uses for DcR3 Antibodies

The DcR3 antibodies of the invention have various utilities. For example, DcR3 antibodies may be used in diagnostic assays for DcR3, e.g., detecting its expression in specific cells, tissues, serum or tumors. Various diagnostic assay techniques known in the art may be used, such as competitive binding assays, direct or indirect sandwich assays and immunoprecipitation assays conducted in either heterogeneous or homogeneous phases [Zola, Monoclonal Antibodies: A Manual of Techniques, CRC Press, Inc. (1987) pp. 147-158]. The antibodies used in the diagnostic assays can be labeled with a detectable moiety. The detectable moiety should be capable of producing, either directly or indirectly, a detectable signal. For example, the detectable moiety may be a radioisotope, such as ^3H , ^{14}C , ^{32}P , ^{35}S , or ^{125}I , a fluorescent or chemiluminescent compound, such as fluorescein isothiocyanate, rhodamine, or luciferin, or an enzyme, such as alkaline phosphatase, beta-galactosidase or horseradish peroxidase. Any method known in the art for conjugating the antibody to the detectable moiety may be employed, including those methods described by Hunter et al., Nature, [144:945] 194:495-496 (1962); David et al., Biochemistry, 13:1014 (1974); Pain et al., J. Immunol. Meth., 40:219 (1981); and Nygren, J. Histochem. and Cytochem., 30:407 (1982). --

On pages 36-37, in the paragraph on lines 34-42 and 1-3, the text has been amended as follows:

--In an alternative technique, DcR3 may be introduced into 293 cells transiently using the dextran sulfate method described by [Somparyrac] Sompayrac et al., Proc. Natl. Acad. Sci., [12] 78:7575 (1981). 293 cells are grown to maximal density in a spinner flask and 700 ug pRK5-DcR3 DNA is added. The cells are first concentrated from the spinner flask by centrifugation and washed with PBS. The DNA-dextran precipitate is incubated on the cell pellet for four hours. The cells are treated with 20% glycerol for 90 seconds, washed with tissue culture medium, and re-introduced into the spinner flask containing tissue culture medium, 5 ug/ml bovine insulin and 0.1 ug/ml bovine transferrin. After about four days, the conditioned media is centrifuged and filtered to remove cells and debris. The sample containing expressed DcR3 can then be concentrated and purified by any selected method, such as dialysis and/or column chromatography. --

On page 38, in the paragraph on lines 18-37, the text has been amended as follows:

--Expressed poly-his tagged DcR3 can then be purified, for example, by Ni^{2+} -chelate affinity chromatography as follows. Extracts are prepared from recombinant virus-infected Sf9 cells as described by [Rupert] Ruppert et al., Nature, 362:175-179 (1993). Briefly, Sf9 cells are washed, resuspended in sonication buffer (25 mL Hepes, pH 7.9; 12.5 mM MgCl_2 ; 0.1 mM EDTA; 10% Glycerol; 0.1% NP-40; 0.4 M KCl), and sonicated twice for 20 seconds on ice. The sonicates are cleared by centrifugation, and the supernatant is diluted 50-fold in loading buffer (50 mM phosphate, 300 mM NaCl, 10% Glycerol, pH 7.8) and filtered through a 0.45 um filter. A Ni^{2+} -NTA agarose column (commercially available from Qiagen) is prepared with a bed volume of 5 mL, washed with 25 mL of water and equilibrated with 25 mL of loading buffer. The filtered cell extract is loaded

onto the column at 0.5 mL per minute. The column is washed to baseline A_{280} with loading buffer, at which point fraction collection is started. Next, the column is washed with a secondary wash buffer (50 mM phosphate; 300 mM NaCl, 10% Glycerol, pH 6.0), which elutes nonspecifically bound protein. After reaching A_{280} baseline again, the column is developed with a 0 to 500 mM Imidazole gradient in the secondary wash buffer. One mL fractions are collected and analyzed by SDS-PAGE and silver staining or western blot with Ni^{2+} -NTA-conjugated to alkaline phosphatase (Qiagen). Fractions containing the eluted His₁₀-tagged DCR3 are pooled and dialyzed against loading buffer. --

On page 42, line 13, the text has been amended as follows:

-- EXAMPLE [12] 11 --

On page 42, line 24, the text has been amended as follows:

-- EXAMPLE [13] 12 --

On page 44, line 31, the text has been amended as follows:

-- EXAMPLE [14] 13 --

On page 47, line 14, the text has been amended as follows:

-- EXAMPLE [15] 14 --

On page 48, line 24, the text has been amended as follows:

-- EXAMPLE [16] 15 --

On page 48, in the paragraph on lines 26-33, the text has been amended as follows:

--An ELISA was conducted to determine if the monoclonal antibodies described in Example [15] 14 were able to bind other known receptors beside DcR3. Specifically, the 4C4.1.4; 11C5.2.8; 8D3.1.5; 5C4.14.7; and 4B7.1.1 antibodies, respectively, were tested for binding to the DcR3 described herein and to DR4 [Pan et al., supra], DR5 [Sheridan et al., supra and Pan et al., supra], DcR1 [Sheridan et al., supra], and OPG [Simonet et al., supra]. The ELISA was performed essentially as described in Example [15] 14 above. Antigen specificity was determined using 10 microgram/ml of DcR3 antibody. --

On page 48, line 38, the text has been amended as follows:

-- EXAMPLE [17] 16 --

On page 49, in the paragraph on lines 5-9, the text has been amended as follows:

--After the washing step, 100 ul of 0.5 ug/ml DcR3 immunoadhesin protein (as described in Example [15] 14 above) or Fas-IgG in assay buffer (PBS containing 0.5% BSA and 0.5% Tween 20) was added to each well. The plates were incubated for 1 hour at room temperature on a shaker apparatus, followed by washing three times with wash buffer. --

On page 49, line 29, the text has been amended as follows:

-- EXAMPLE [18] 17 --

On pages 49-50, in the paragraph on lines 31-42 and line 1, the text has been amended as follows:

--The isotype of the DcR3 antibodies (as described above in Examples [15-17] 14-16) was determined by coating microtiter plates with isotype specific goat anti-mouse Ig (Fisher Biotech,

Pittsburgh, PA) overnight at 4°C. The plates were then washed with wash buffer (as described in Example [15] 14 above). The wells in the microtiter plates were then blocked with 200 ul of 2% bovine serum albumin (BSA) and incubated at room temperature for one hour.

The plates were washed again three times with wash buffer. Next, 100 ul of hybridoma culture supernatant or 5 ug/ml of purified antibody was added to designated wells. The plates were incubated at room temperature for 30 minutes and then 50 ul HRP-conjugated goat anti-mouse IgG (as described above in Example [15] 14) was added to each well. The plates were incubated for 30 minutes at room temperature. The level of HRP bound to the plate was detected using HRP substrate as described above. --

On page 51, in the paragraph on lines 2-16, the text has been amended as follows:

-- Deposit of Material

The following materials have been deposited with the American Type Culture Collection, 10801 University Blvd., Manassas, Virginia USA (ATCC):

<u>Material</u>	<u>ATCC Dep. No.</u>	<u>Deposit Date</u>
DNA30942-1134	209254	Sept. 16, 1997
4C4.1.4	<u>HB-12573</u>	Sept. <u>18</u> , 1998
5C4.14.7	<u>HB-12574</u>	Sept. <u>18</u> , 1998
11C5.2.8	<u>HB-12572</u>	Sept. <u>18</u> , 1998
8D3.1.5	<u>HB-12571</u>	Sept. <u>18</u> , 1998
4B7.1.1	<u>HB-12575</u>	Sept. <u>18</u> , 1998

Claims 1, 7, and 14-29 have been canceled without prejudice.

Claims 30-39 have been amended as follows:

30. (Amended) The hybridoma cell line 4C4.1.4 deposited as ATCC accession number [____] HB-12573.

31. (Amended) The hybridoma cell line 5C4.14.7 deposited as ATCC

accession number [____] HB-12574.

32. (Amended) The hybridoma cell line 11C5.2.8 deposited as ATCC accession number [____] HB-12572.

33. (Amended) The hybridoma cell line 8D3.1.5 deposited as ATCC accession number [____] HB-12571.

34. (Amended) The hybridoma cell line 4B7.1.1 deposited as ATCC accession number [____] HB-12575.

35. (Amended) The 4C4.1.4 monoclonal antibody produced by the hybridoma cell line deposited as ATCC accession number [____] HB-12573.

36. (Amended) The 5C4.14.7 monoclonal antibody produced by the hybridoma cell line deposited as ATCC accession number [____] HB-12574.

37. (Amended) The 11C5.2.8 monoclonal antibody produced by the hybridoma cell line deposited as ATCC accession number [____] HB-12572.

38. (Amended) The 8D3.1.5 monoclonal antibody produced by the hybridoma cell line deposited as ATCC accession number [____] HB-12571.

39. (Amended) The 4B7.1.1 monoclonal antibody produced by the hybridoma cell line deposited as ATCC accession number [____] HB-12575.

The following Claims 67-89 are being added:

--67. An isolated anti-DcR3 antibody which (a) specifically binds to a DcR3 polypeptide consisting of amino acids 1 to 215 or amino acids 1 to 300 of Fig. 1 (SEQ ID NO:1) and (b) inhibits binding of

Fas ligand to the DcR3 polypeptide of (a).

68. The antibody of claim 67 wherein said antibody is a monoclonal antibody.

69. The antibody of claim 68 wherein said antibody is a chimeric antibody.

70. The antibody of claim 68 wherein said antibody comprises a Fab fragment.

71. The antibody of claim 68 wherein said antibody is a monovalent antibody.

72. The antibody of claim 68 wherein said antibody is a human antibody.

73. The antibody of claim 68 wherein said antibody is expressed in a recombinant host cell selected from the group consisting of a CHO cell, yeast cell and *E. coli*.

74. The antibody of claim 67 wherein said antibody further inhibits binding of LIGHT ligand to the DcR3 polypeptide of (a).

75. An isolated anti-DcR3 antagonist antibody which (a) specifically binds to a DcR3 polypeptide consisting of amino acids 1 to 215 or amino acids 1 to 300 of Fig. 1 (SEQ ID NO:1), (b) blocks binding of Fas ligand to the DcR3 polypeptide of (a), and (c) neutralizes inhibitory effect of the DcR3 polypeptide of (a) on Fas ligand activity in one or more mammalian cells.

76. The antibody of claim 75 wherein said antibody neutralizes inhibitory effect of the DcR3 polypeptide of (a) on Fas ligand-induced apoptosis in one or more mammalian cells.

77. The antibody of claim 76 wherein said antibody neutralizes inhibitory effect of the DcR3 polypeptide of (a) on Fas ligand-induced apoptosis in one or more mammalian cancer cells.

78. The antibody of claim 77 wherein said mammalian cancer cells are colon cancer cells or lung cancer cells.

79. The antibody of claim 75 wherein said antibody is a monoclonal antibody.

80. The antibody of claim 79 wherein said antibody is a chimeric antibody.

81. The antibody of claim 79 wherein said antibody comprises a Fab fragment.

82. The antibody of claim 79 wherein said antibody is a monovalent antibody.

83. The antibody of claim 79 wherein said antibody is a human antibody.

84. The antibody of claim 79 wherein said antibody is expressed in a recombinant host cell selected from the group consisting of a CHO cell, yeast cell and *E. coli*.

85. An isolated anti-DcR3 antibody which specifically binds to a DcR3 polypeptide comprising amino acids 1 to 215 of Fig. 1 (SEQ ID NO:1) and binds to the same DcR3 polypeptide epitope as the epitope to which the 4C4.1.4 monoclonal antibody produced by the hybridoma cell line deposited as ATCC accession number HB-12573 binds.

86. An isolated anti-DcR3 antibody which specifically binds to a DcR3 polypeptide comprising amino acids 1 to 215 of Fig. 1 (SEQ ID

NO:1) and binds to the same DcR3 polypeptide epitope as the epitope to which the 5C4.14.7 monoclonal antibody produced by the hybridoma cell line deposited as ATCC accession number HB-12574 binds.

87. An isolated anti-DcR3 antibody which specifically binds to a DcR3 polypeptide comprising amino acids 1 to 215 of Fig. 1 (SEQ ID NO:1) and binds to the same DcR3 polypeptide epitope as the epitope to which the 11C5.2.8 monoclonal antibody produced by the hybridoma cell line deposited as ATCC accession number HB-12572 binds.

88. An isolated anti-DcR3 antibody which specifically binds to a DcR3 polypeptide comprising amino acids 1 to 215 of Fig. 1 (SEQ ID NO:1) and binds to the same DcR3 polypeptide epitope as the epitope to which the 8D3.1.5 monoclonal antibody produced by the hybridoma cell line deposited as ATCC accession number HB-12571 binds.

89. An isolated anti-DcR3 antibody which specifically binds to a DcR3 polypeptide comprising amino acids 1 to 215 of Fig. 1 (SEQ ID NO:1) and binds to the same DcR3 polypeptide epitope as the epitope to which the 4B7.1.1 monoclonal antibody produced by the hybridoma cell line deposited as ATCC accession number HB-12575 binds. --